

Patient gender as a factor associated with lymph node metastasis in T1 colorectal cancer: A systematic review and meta-analysis

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Abstract. Approximately 10% of patients with T1 colorectal cancer have lymph node metastases (LNM), requiring node dissection along with surgical resection. Patient gender was recently reported to affect the occurrence of LNM. The aim of the present study was to assess whether patient gender was predictive of LNM in T1 colorectal cancer. Public databases, including PubMed, EMBASE and the Cochrane Central Register of Controlled Trials were searched, using key terms related to 'T1 colorectal cancer' and 'lymph node'. All relevant studies reporting the adjusted odds ratio or risk ratio of LNM in relation to patient gender were included. The quality of the studies was classified according to the Quality in Prognostic Studies tool. A random-effects model was used and the quality of the evidence was evaluated using the Grading of Recommendations Assessment, Development and Evaluation approach. The initial database search identified 2,492 publications; of those, 36 studies reported unadjusted results. Of the 36 studies, 4 reported adjusted results and fulfilled the inclusion criteria for this meta-analysis: 3 studies were graded as having a moderate risk of bias, and 1 had a low risk of bias. The present meta-analysis demonstrated that female gender was associated with increased risk of LNM (risk ratio=2.45, 95% confidence interval: 1.03-3.88). The I² statistic was 0.901, classified as very low (+000) and was downgraded by the risk of bias, inconsistency and publication bias. In conclusion, female gender was found to be correlated with LNM in patients with T1 colorectal cancer.

Introduction

Colorectal cancer is one of the most common types of cancer worldwide. Due to the advances in endoscopic treatment, particularly endoscopic submucosal dissection, several T1 colorectal cancers are resected endoscopically with negative margins (1-3). Lymph node metastasis (LNM) occurs in ~10% of patients with T1 colorectal cancer, with these patients requiring surgical resection with lymph node dissection (4-7). Therefore, determining risk factors associated with LNM in patients with T1 colorectal cancer is crucial.

A number of studies have assessed factors predictive of LNM in patients with T1 colorectal cancer. Previously identified risk factors for LNM include lymphovascular invasion, histological grade, tumor budding and degree of submucosal invasion (8-10). These factors are included in various diagnostic and treatment guidelines, including those of the National Comprehensive Cancer Network, the European Society for Medical Oncology and the Japanese Society for Cancer of the Colon and Rectum (8-10). However, the majority of the studies identifying these guidelines were retrospective in design and included small numbers of patients. In addition, these analyses were limited to pathological factors. The indications for additional surgery plus lymph node dissection following endoscopic resection remain unclear.

A recent retrospective, single-center study, which included a large number of patients, reported that female gender was associated with LNM in patients with T1 colorectal cancer (4). Other studies also reported higher rates of LNM in female compared with male patients, although these differences were not statistically significant (11,12). Several systematic reviews and meta-analyses have investigated risk factors for LNM; however, none has focused on patient gender as a predictive factor for LNM to date (13-17). The aim of the present systematic review and meta-analysis was to assess whether the gender of patients with T1 colorectal cancer is predictive of LNM.

Materials and methods

Search strategy and selection criteria. This systematic review and meta-analysis was performed according to the Preferred

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Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) statement, was conducted in accordance with the Cochrane Handbook (18,19) and was pre-registered (CRD42015024588). MEDLINE, EMBASE and the Cochrane Central Register of Controlled Trials were searched from the earliest date of indexing through July 11, 2015. The search terms included 'T1', 'early', 'colorectal', 'colonic', 'rectal', 'adenocarcinoma', 'neoplasm', 'lymph node', 'N1' and 'N2' in various combinations. Additional searches were performed by manual cross-referencing. Only studies published in English were included. The meta-analysis was restricted to studies reporting the adjusted odds ratio (aOR) or risk ratio (RR) of dissection-diagnosed LNM in relation to gender in patients with T1 colorectal cancer. Patients with familial adenomatous polyposis, Lynch syndrome and ulcerative colitis were excluded, as were patients who underwent only endoscopic treatment or transanal endoscopic microsurgery.

Data extraction. Two authors (K.I. and Y.K.) independently reviewed the abstracts and titles identified by the searches. All studies rated as possible candidates by either of these two reviewers were included in the preliminary list, and their full texts were retrieved. The two authors independently reviewed the full texts to determine whether the studies met the review criteria. Disagreements were resolved by discussion, or if necessary by a third reviewer (Y.K.). Information extracted from studies deemed to have met the review criteria included name of first author, year of publication, country, study design, number of patients, study inclusion and exclusion criteria, study quality, demographic data and outcome events.

Risk of bias. Two authors (K.I. and Y.K.) independently assessed the risk of bias using the Quality in Prognostic Studies (QUIPS) tool (20). Each domain was rated as being at low, high or unclear risk of bias, based on whether the study sample adequately represented the population of interest; whether the participants not lost to follow-up adequately represented the study sample; whether prognostic factors and outcomes of interest were measured similarly for all participants; and whether there were other sources of bias. Disagreements between reviewers were resolved by discussion.

Statistical analysis. Data were analyzed by a single investigator (Y.K.). All studies included in the meta-analysis reported the frequency of LNM in men and women, either in the text or in the tables. Data were synthesized using Stata software, version 13.0 (Stata Corp., College Station, TX, USA). A meta-analysis was performed to summarize the prognostic effects of gender, with results reported as RR and 95% confidence interval (CI). A random-effects model was used. The quality of evidence was evaluated using the Grading of Recommendations Assessment, Development and Evaluation (GRADE) approach (21). Heterogeneity was assessed by visual inspection of the forest plots. I^2 statistics were calculated and analyzed based on the recommendations of the Cochrane Handbook, in which I^2 values of 0-40, 30-60, 50-90 and 75-100% represent little, moderate, substantial and considerable heterogeneity, respectively (19). A sensitivity analysis was conducted to pool all 36 studies reporting unadjusted relative risk of gender. A subgroup analysis could not be conducted

due to data insufficiency. P-values <0.05 were considered to indicate statistically significant differences.

Results

Study selection and inclusion. The initial database search identified 2,492 publications. Following removal of duplicates, 2,489 unique publications were identified, 1,419 on PubMed, 1,889 on EMBASE and 162 on the Cochrane Library. Three additional publications were identified through other sources or from the references lists of the included publications. After screening the titles and abstracts, 441 full-text articles were assessed for eligibility (first review). Of those, 36 studies reported unadjusted results and were included for systematic review (second review). Of the 36 studies, 4 (4,22-24) reported adjusted results and fulfilled the predetermined inclusion criteria for the meta-analysis (Fig. 1).

Characteristics of included studies. The 4 studies included in this meta-analysis were retrospective in design and involved 1,329 patients with T1 colorectal cancer. Of the 4 studies, 3 were single-center and 1 was a multicenter study. As regards bias, 3 studies were graded as having a moderate risk of bias and 1 as having a low risk of bias. The median number of patients per study was 332 (range, 142-653). Of the 1,329 included patients, 864 (65.0%) were male and 465 (35.0%) were female; 558 (42.0%) had rectal carcinomas and 771 (58.0%) had colon carcinomas. The characteristics of the 4 included studies are presented in Table I.

Of the 1,329 patients, 113 (8.5%; 95% CI: 7.1-10.1) were positive for LNM, with the number per study ranging from 6.3 to 9.9%. The incidence of LNM was 6.4% (55/864, 95% CI: 4.8-8.2) in male and 12.5% (58/465, 95% CI: 9.6-15.8) in female patients.

Quality of evidence. Publication bias could not be evaluated using funnel plots or Egger's regression test. Only 4 of 36 studies reported adjusted outcomes, suggesting a selective outcome reporting bias (25). The risk of bias was serious, as the number of studies with a low risk of bias was limited. The I^2 statistic was 0.901, classified as very low (+000), and was downgraded by the risk of bias, inconsistency and publication bias (Table II).

Patient gender as a predictive factor for LNM. Of the 4 studies, 3 reported a higher rate of LNM in female compared with male patients with T1 colorectal cancer (10.8 vs. 4.6%, 15.7 vs. 4.2% and 12.7 vs. 7.1%, respectively), whereas 1 study reported a lower rate of LNM in female patients (8.3 vs. 10.6%) (4,22-24). Of the 4 studies, 2 (4,23) reported that female gender was an independent risk factor for LNM in patients with T1 colorectal cancer (OR=5.68 and 2.22, respectively), whereas the remaining 2 studies (22,24) reported no significant difference between male and female patients on the univariate as well as the multivariate analyses. The result of the meta-analysis for multivariate risk ratio is shown in Fig. 2. The weights were from the random-effects analysis. The meta-analysis demonstrated that female gender was associated with LNM in patients with T1 colorectal cancer (RR=2.45, 95% CI: 1.03-3.88).

Sensitivity analysis. The pooled sensitivity analysis of the 36 studies revealed that female gender was associated with LNM

Table I. Characteristics of included studies.

First author, year	Country	Number of patients (women)	Risk of bias					Statistical analysis and reporting (Refs.)
			Study participation	Study attrition	Prognostic factor measurement	Outcome measurement	Study confounding	
Yamamoto, 2004	Japan	301 (83)	Low	Low	Low	Moderate	Low	Low (24)
Kobayashi, 2010	Japan	233 (89)	Low	Low	Low	Unclear	Low	Low (23)
Umemura, 2013	Japan	142 (48)	Moderate	Low	Low	Moderate	Low	Low (22)
Miyachi, 2015	Japan	653 (245)	Low	Low	Low	Low	Low	Low (4)

Table II. GRADE evidence profile.

No. of studies reporting adjusted results on node positivity	Quality assessment				Effect		
	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations	Proportion (95% CI)
4	Observational	Serious ^a	Serious ^b	Not serious	Not serious	Publication bias strongly suspected ^c	Increase event proportion 35.7/1,000 individuals (0.74-71.0)
							+ OOO (very low ^{a-c})
							Important

^aFew studies exhibited low risk of bias. ^bI²=90.1%. ^cOf the 36 studies, 4 reported adjusted outcomes for gender. GRADE, Grading of Recommendations Assessment, Development and Evaluation; CI, confidence interval.

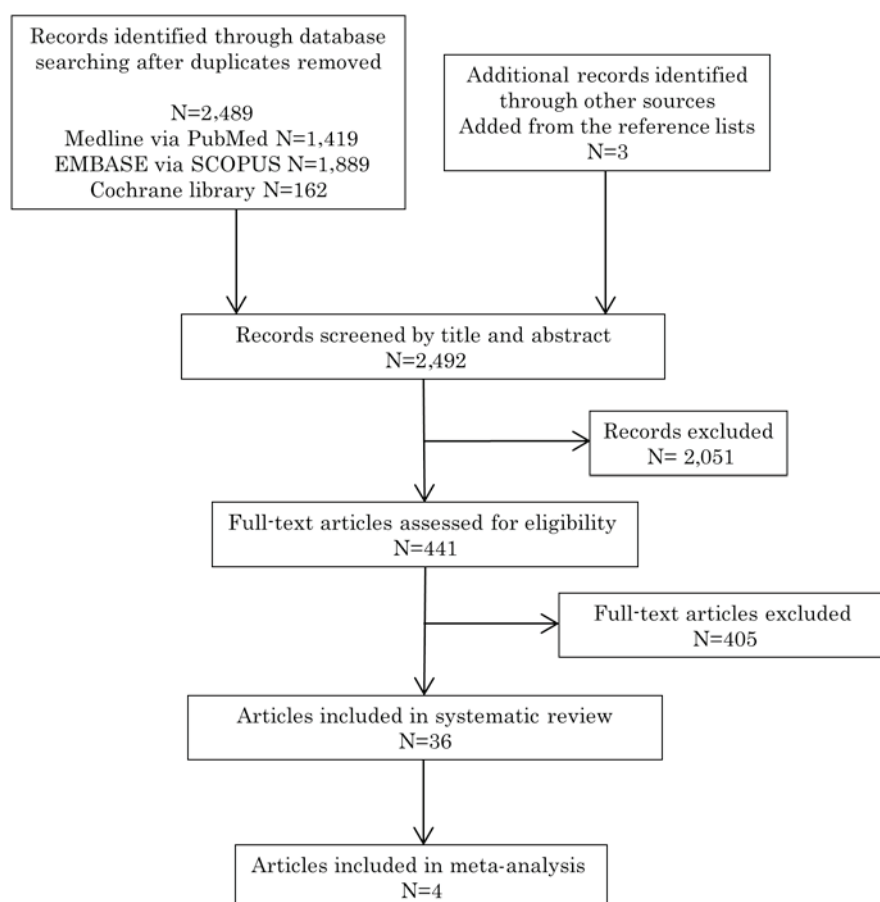


Figure 1. Flow diagram of the study selection process.

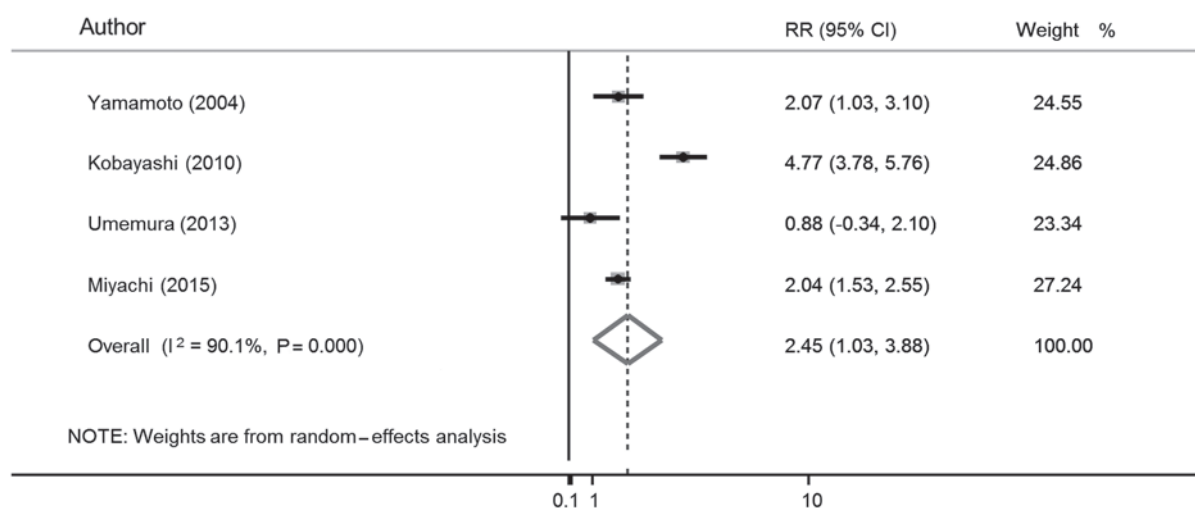


Figure 2. Meta-analysis for multivariate risk ratios (RRs). CI, confidence interval.

(RR=1.33, 95% CI: 1.17-1.51; Fig. 3), which was consistent with the main results (4-6,11,12,22-24,26-49).

Discussion

In the present study, the association between patient gender and LNM in patients with T1 colorectal cancer was systematically reviewed. Our meta-analysis revealed that female gender

was associated with LNM in T1 colorectal cancer. To the best of our knowledge, this is the first such analysis showing that patient gender is predictive of LNM in patients with T1 colorectal cancer.

Overall, ~10% of patients with T1 colorectal cancers have LNM, thereby requiring more invasive surgery along with lymph node dissection (4-7). Operative treatments are relatively invasive and costly, making local excision an attractive

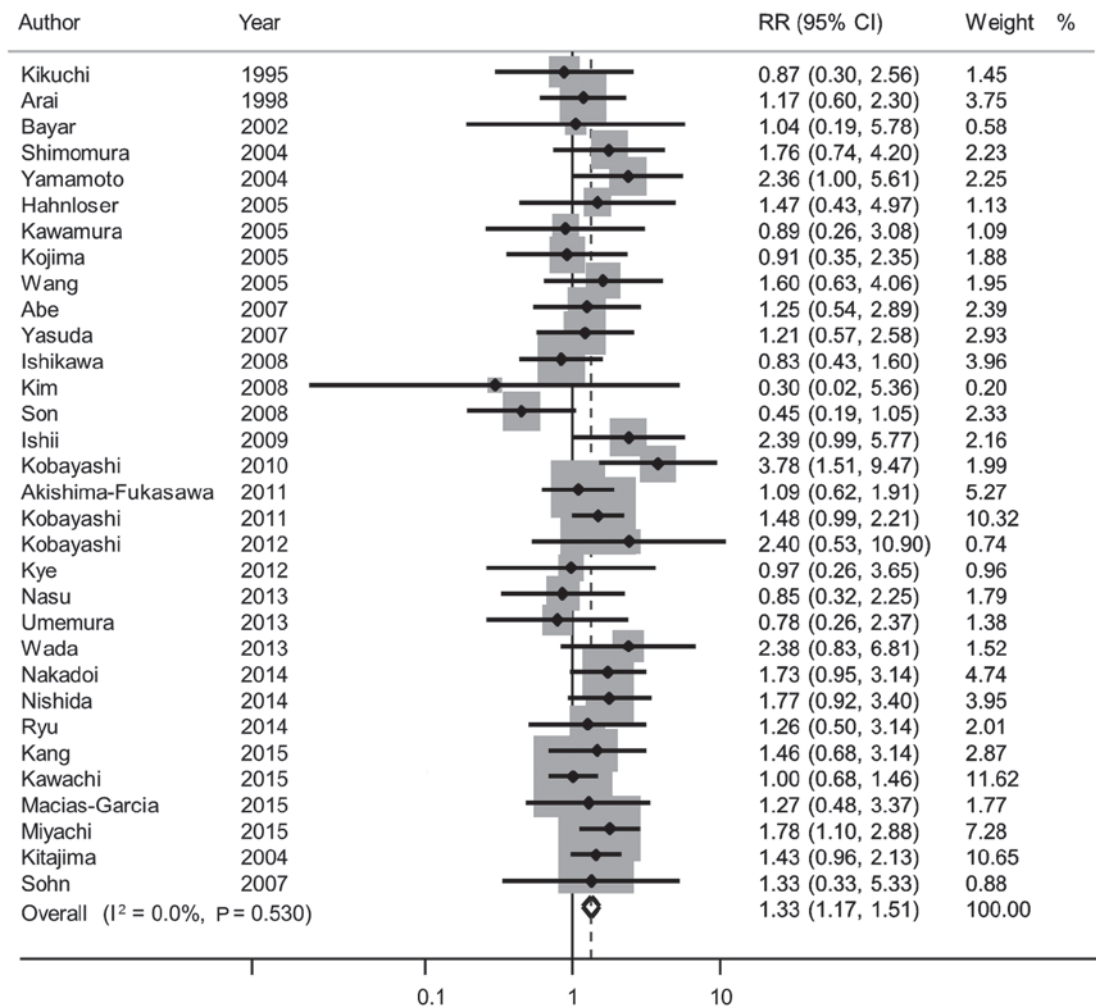


Figure 3. Sensitivity analysis. RR, risk ratio; CI, confidence interval.

treatment option. However, local excision is oncologically safe only in the absence of LNM. As LNM is difficult to assess preoperatively, the decision to perform radical surgery following endoscopic resection is based on the results of clinicopathological analysis. Several previous systematic reviews of small, retrospective studies have identified reliable pathological factors associated with the risk of LNM in T1 colorectal cancer (13-17). These meta-analyses reported that depth of submucosal invasion $>1,000 \mu\text{m}$, lymphovascular invasion, poorly differentiated tumors and tumor budding were all risk factors for LNM. The diagnosis of pathological factors may differ among observers (38,50). Moreover, pathological diagnoses may depend on the immunohistochemical assay used, such as D2-40, Victoria Blue and CAM 5.2. For example, lymphatic invasion is more accurately diagnosed using an anti-human podoplanin antibody rather than by hematoxylin and eosin staining (51-53). By contrast, our meta-analysis was the first to demonstrate that patient gender as a new clinical risk factor was predictive of LNM. Moreover, in contrast to the other meta-analyses, ours assessed the risk of bias of each study using the QUIPS tool and evaluated the quality of evidence using the GRADE approach.

A recent study of 653 patients with T1 colorectal cancer demonstrated that female gender was an independent risk factor for LNM (4). Stratification of patients according to the

status of the muscularis mucosae (whether the muscle fibers were maintained or fragmented/disappeared), pathological factors and patient gender provides more appropriate indications for additional surgery along with lymph node dissection in this patient population, and may help reduce the incidence of unnecessary surgery. Several other studies also reported that the rate of LNM was higher in female compared with male patients (5,11), and that female gender was an independent risk factor for LNM in patients with T1 lower rectal cancer (23).

Although the mechanism underlying the higher rate of LNM in women with T1 colorectal cancer has not been fully elucidated, epidemiological studies have reported a potential association between gender hormones and colorectal cancer (54-56). Some studies indicate a role for estrogen in the protection against colorectal cancer (55-58). The effects of estrogen are mediated by estrogen receptors (ERs), namely ER α and ER β (59,60). ER β expression was found to be significantly reduced in adenomatous tissues with high levels of dysplasia as well as in carcinomatous tissues compared with normal mucosa (61). In addition, the degree of ER β expression loss appears to be correlated with more advanced stage and higher tumor grade (62,63). The degree of reduction in ER β level may also be correlated with LNM. Nussler *et al* reported that ER β levels were significantly reduced in colorectal cancer in both men and women compared with normal colonic mucosa, and this reduction in ER β level

was different by gender (64). Other studies were unable to detect such gender differences (62,65). However, the samples of all those studies were very limited and investigation using larger sample sizes would be required to demonstrate the difference in ER β levels by gender. Furthermore, not only a reduction of the ER β levels, but more importantly, a change in the ER α :ER β ratio, may determine the susceptibility of a given tissue to carcinogenesis (66,67). This is only one plausible reason and there may be other possible explanations for the association between LNM and gender in T1 colorectal cancer; therefore, further investigation is required.

This meta-analysis had several limitations. The main limitation was the lack of randomized controlled trials, as confounding factors may be more effectively removed from a randomized trial rather than from an observational study. As the patients in these studies were not randomized by gender, our analysis may have been sensitive to confounding variables. Therefore, only studies with adjusted results were included. Second, only 4 of the 36 studies reported adjusted results. Thus, a selective outcome reporting bias may have led to the gender-related difference in LNM rate. Our sensitivity analysis included all 36 studies, with the results not differing markedly. Third, all the studies in this meta-analysis originated in Japan, which may have affected our results. Only 3 of the 36 studies were from western countries, none of which reported adjusted results, and were thus excluded from the current criteria (28,30,48). In fact, these 3 studies showed a tendency of higher LNM rate in female rather than in male patients, but the difference was not significant due to insufficient number (<100) of study subjects. The association between female gender and LNM may differ by race. However, such a meta-analysis including western populations cannot be conducted at present; thus, this risk factor requires larger-scale validation in western countries. In our meta-analysis, 1 of the 4 included studies reported a higher LNM rate in male rather than female patients, although the difference was not statistically significant. There was little clinical heterogeneity. Thus, this difference may be due to chance by small sample size.

In conclusion, the gender of patients with T1 colorectal cancer was found to be predictive of LNM. This finding may help select patients who may be spared radical resection, thereby preventing unnecessary surgery without compromising oncological safety. Further prospective randomized studies with larger patient populations are required to confirm this result.

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